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Antidepressant, Antidiarrheal, Thrombolytic and Phytochemical Profiling of Ethanol Extract of *Amomum aromaticum* Leaves: *In vivo* and *In vitro* Approaches

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Abstract

The study evaluated several pharmacological effects including antidepressant, antidiarrheal and thrombolytic activities of ethanol extract of *Amomum aromaticum* leaves (EEAA). Upon the assessment for qualitative phytochemical groups in the extract, several doses of EEAA (1000 - 4000 mg/kg) were studied to find acute oral toxicity in mouse for safe dose selection. Then EEAA was tested whether it demonstrates antidepressant activity in tail suspension test (TST) and forced swim test (FST). Anti-diarrheal and clot lysis activities of EEAA were evaluated in castor oil-induced diarrhoea in mouse and *In vitro* clot lysis method, respectively. Oral administration of EEAA (1000 - 4000 mg/kg) showed no morality after 10 days, and no sign of acute toxicity observed within 24 hrs post-treatment. The qualitative phytochemical screening showed the presence of carbohydrate, alkaloid, flavonoid, tannin, saponin, and polyphenol groups in EEAA. The TST and FST resulted with significant improvement in mobility in mice treated with EEAA (400 mg/kg), where fluoxetine (20 mg/kg) was used as standard in both tests. EEAA treatment also showed a moderate dose-dependent anti-diarrheal effect. The 400 mg/kg oral dosing for 14 days decreased the rate of defecation by 52.8% compared to the control group. This study also demonstrated that EEAA possesses clot lysis activity. Hence, further intense investigations are suggested to identify specific potential active phytochemicals.

Key words: *Amomum aromaticum,* ethanol extract, anti-depressants, anti-diarrheal, thrombolytic.

Introduction

Depression is the most common neurological disorder in the world that could affect any age population. Etiology of depression includes loss of appetite, lack of energy, sleep deprivation or sometimes rapid eye movement sleep, leading to suicide attempt (Martins and Brijesh S 2018). Various anti-depressant drugs are being prescribed frequently to treat depression, but their complications in long-term use made researchers find new leads from safe and available sources.

A high portion of electrolytes is removed from the body on diarrhea, making patient stressed, weak and dismantle systemic homeostasis. Diarrhea is one leading cause of child mortality in developing countries (Toyin *et al.*, 2012). Although healthy sanitation has assured diarrhea prevention in several countries, infectious diarrhea by a virus and bacterial invasion is inevitable (Gakunga *et al.*, 2013). There are several anti-diarrheal drugs available in the market, including loperamide

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hydrochloride, bismuth subsalicylateetc. These drugs have some common side-effects like abdominal pain, nausea, vomiting, anxiety anal discomfort and constipation (Johnson *et al*., 1986). Therefore, urging a safe and drug source and herbs is one mostly researched source for antidiarrheal drugs.

Thrombolysis is a process of dissolving blood coagulation where thrombolytic agents activate plasminogen and cleave to form plasmin. Plasmin, a proteolytic enzyme, breaks structural integrity of clot by breaking the fibrin matrix. Endogenous blood coagulation hinders blood supply harmony that may cause mild to severe cardiovascular diseases like myocardial infection and stroke (Hossain *et al.*, 2019).

A study has found that 80% of the world's people somehow depend on these herbal and traditional medicinal plants (Khan *et al*., 2011). Typically, certain chemical groups present in the extract of a plant can cure certain diseases such as diarrhea, depression, diabetics, anxiety, thrombosis, etc. (Awuchi, 2019). *Amomum aromaticum* Roxb. is one of the known traditional medicines in Bangladesh that has been used in the treatment of diarrhea and depression. *A. aromaticum* is locally known as "Bengal Cardamon". This plant is also used as a folk medicine to treat joint pains (Sharma *et al*., 2004), fever, abortifacient (Kala, 2005), vomiting, cough, inflammation and malaria (Dang *et al*., 2020). Anti-depressants, anti-diarrheal and thrombolytic effects of plant extracts are related to phytochemical groups such as alkaloids, terpenes and terpenoids, saponins and sapogenins, amines, carbohydrates, flavonoids, polyphenols and tannins (Bahramsoltani *et al*., 2015; Ghosh *et al*., 2015; Rahman *et al*., 2015). The potentials of *A. aromaticum* against depression, diarrhea and thrombolysis are yet to be investigated. In the present study, the ethanol extract of *A. aromaticum* leaves (EEAA) was investigated using *in vitro* and *in vivo* models for the said pharmacological activities.

Materials and Method

Extract preparation: Fresh leaves of *A. aromaticum* (identified by taxonomist Prof. Shaikh Bokhtear Uddin, University of Chittagong, Chattogram, Bangladesh) were collected from the Chittagong hill tract area. The collected leaves were washed with water and dried at room temperature under air for 12 days. Dried leaves were kept at 60-70^0 C for 2 hours for further drying. Leaves were ground to get coarse powder by the help of a mechanical grinder. About 500 gm powdered materials were soaked in 1L of ethanol in a closed glass bottle for 10 days with consistent shaking. The dark greenish solution was filtered through Buchner funnel, and residual solvents were evaporated to obtain final greasy and semisolid EEAA extract, which was stocked at 4^{o}C until further use (Islam *et al*., 2017).

Drugs and chemicals: Fluoxetine, loperamide, and streptokinase were purchased from Square Pharmaceuticals Ltd., Bangladesh. Ethanol and sodium hydroxide were procured from the Merck (Darmstadt, Germany). Chloroform and ferric chloride were purchased from the Hindon India Pvt. Ltd., and glacial acetic acid was collected from the Sigma chemicals co. (St. Louis, MO).

Animals: Male *Swiss albino* mice (~25 gm) were purchased from Bangladesh Council of Scientific and Industrial Research (BCSIR), Chittagong, Bangladesh. They were housed one week for acclimatisation under 12 h day-night circadian rhythm and supplied with *ad libitum* standard rodent chaw and water. All experiments were conducted at room temperature and laboratory condition. The experimental protocol was approved by the Planning and Development Committee, Department of Pharmacy, International Islamic University Chittagong, Chattogram, Bangladesh (Pharm-P&D-37/07'12). At the end of each experiment, animals were killed by cervical dislocation and disposed of following standard procedure.

Phytochemical analysis: EEAA was subjected to qualitative phytochemical analysis to confirm its phytochemicals such as alkaloids, flavonoids,

carbohydrates, tannins, phenols, glycosides, proteins, saponins, terpenoids and mucilages etc. followed by a slight modification of Trease and Evans (1989), Sofowara (1993) and Harborne (1973) (Akinmoladun *et al*., 2007; Awoyinka *et al*., 2007).

Acute toxicity: Acute toxicity study was done on twenty *Swiss albino* mice dosing 1000 mg/kg, 2000 mg/kg, 3000 mg/kg and 4000 mg/kg and administered orally by gavage. Observations were performed closely for the next 10 days and checked out the alteration of behavioural activity and mortality (Ganapaty *et al*., 2002).

Tail suspension test: The tail suspension test (TST) is a method developed by Steru *et al*. 1982 (Steru *et al*, 1985) to screen potential anti-depressant drugs and assess activity in depression-related behaviour in a rodent model. Briefly, mice were divided into four groups (Control, Standard, EEAA 200 mg/kg and EEAA 400 mg/kg); each group containing 5 mice. Cleaned tails of mice were wrapped with adhesive tapes at half distance from the base, and an experimental clip was attached to that tape. Mice were suspended from 50 cm above the floor and observed for the immobility of animals like lack of attempt to move limbs or being idle in a vertical posture. The observer used a stopwatch to record time till immobility and full experiment period were 6 min.

Forced swimming test: Forced swimming test was developed to screen anti-depressant drugs, method following a previously described (Arauchi *et al*., 2018). Animals were separated into 4 groups; each animal from different groups was placed into a plastic cylinder (25 cm × 15 cm × 25 cm). The vessel was filled with water (10 L; depth 40 cm), and temperature was maintained at 25±1°C. On preparing session, individual mice could swim for 10 min for acclimation, during acquisition the session shortened to 6 min. Mouse remained motionless, floating, and rare movement only to keep heads above water were considered as immobility (Zomkowski *et al*., 2005; Zomkowski *et al*., 2004).

Castor oil-induced diarrhea in mice: To measure the anti-diarrheal activity mice were starved for 16 hr before the experiment. On the day of experiment, animals subjected to induce laxation were fed 0.5 ml of castor oil orally; control group mice received saline only, and rest of group mice received respective doses as prescribed (Shoba and Thomas, 2001; Tunaru, 2012). Post treatment animals were placed in clear cage, floor covered by white tissue paper. The shapeless and watery stools were measured hourly for a total 4 hr period. The percent of inhibition of defecation was calculated by the following equation-

$$Inhibition\ of\ defecation\ (\%) = \frac{A-B}{A} \times 100$$

Where, A = mean number of defecation feces of the control group and B = mean number of defecation caused by plant extract.

In vitro thrombolytic activity: Blood clot dissolution test of EEAA was conducted by minor alteration of Prasad *et al*. (2007) (Dewan & Das, 2013). Healthy volunteers were selected based on several screening process including no drug record in last six months, no record of cardiac dysfunction, blood pressure stable during sampling and not diagnosed for any infectious disease, and non-alcoholic. Four ml blood was withdrawn from median cephalic vein and preserved in sodium citrate tube until use in experiment. In the experiment, 0.5 ml of blood was taken into pre-weighed micro-centrifuge tubes and mixed with 200 µl of 1% calcium chloride; the mixture was incubated at 37° C for 45 minutes, for clot formation. Once clot formed, serum was sucked off completely and weighed each tube again to ascertain the clot weight.

Streptokinase (100 µl) and plant extract at prescribed doses were added to each tube, while the control group was filled with similar volume of distilled water. The mixture incubated at 37^0 C for 90 minutes, remaining fluids were removed without disturbing pellets at the bottom. Final weight measured, and the percent of clot lysis was calculated by the following equation-

$$\%\ of\ clot\ lysis = \frac{wt.of\ lyses}{wt.of\ clot}\ X\ 100$$

Results and Discussion

The present study has revealed three major pharmacological activities of EEAA. Firstly, the immobility time was significantly shorter in EEAA treated mice than control group, suggesting reduced depression-like behaviour. Second, extract treatment showed significant reduction in watery pooping-induced by castor oil. Finally, we showed EEAA have great potentiality to activate plasminogen enzyme, thus, can break endogenous thrombi and protect from blood clot related diseases.

Acute toxicity: Acute toxicity was observed for 10 days but no alteration were shown by following oral administration of EEAA (1000 mg/kg to 4000 mg/kg). As a result, dosage levels of 200 and 400 mg/kg were used for the current study.

Qualitative phytochemical analysis: The primary phytochemical study of EEAA exhibited the presence of several phytochemicals like carbohydrates, alkaloids, flavonoids, tannins, saponins, and polyphenols (Table 1).

Table 1. Qualitative phytochemical screening of EEAA.

Phytochemicals	Appearance	Results
Alkaloids	Mayer's test: Yellow colour Wagner test: A reddish-brown colour	+
Carbo-hydrates	Reddish colour ring form	+
Glycosides	No ring form	_
Reducing sugar	Fehling's test: No colour form Benedict's test: Reddish colour precipitate form	_
Flavonoids	Florescence yellow colour form	+
Saponins	Persistent forth for one hour	+
Tannin	Brownish blue appears	+
Polyphenol	Violet colour form	+
Resin	No precipitation	_
Terpenoids	Reddish-brown not form	_

Signs (+) and (-) indicate the presence and absence of phytochemical classes.

In vivo anti-depressant activity: The anti-depressants activities of EEAA was evaluated using stressful and inescapable conditions, tail suspension and forced swimming test. In both tests, immobility time was the indicator of giving up situation and learning helplessness, which are the sign of depression like behavior (Cryan and Mombereau, 2004). In tail suspension test, the mean immobility time after EEAA (400 mg/kg) treatment was 75 ± 2 s, which was significantly (p<0.01; n=6) shorter than control group (180.67 ± 1.10 s) (Figure 1). Fluoxetine was used as standard and exhibited significant (p<0.001) shorter immobility time (69.87 ± 1.45 s) that validating the study. To further confirm the anti-depressant activity of EEAA (200 and 400 mg/kg), forced swim test was used. Like the tail suspension test, EEAA (400 mg/kg) produced a moderate reduction in immobility time (67.667±2.45s) than control (Figure 2).

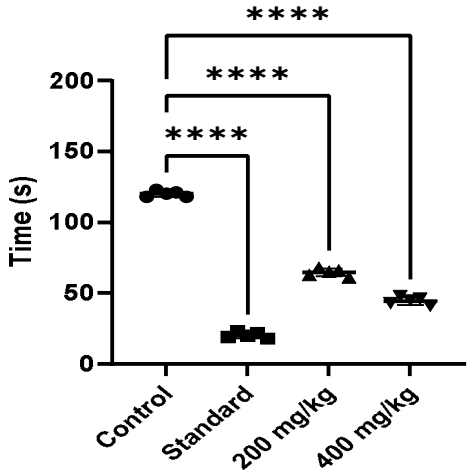

Figure 1. *In vivo* force swimming test (FST) of ethanol extract of *Amomum aromaticum* leaves. Values are expressed as mean ± SEM (n = 5); ****p< 0.001 are statistically significant compared to control followed by Dunnet test (GraphPad Prism 8.4). (EEAA) = Ethanol extract of *Amomum aromaticum*.

Anti-diarrheal activity: Castor oil is one of the oldest drugs, which cause laxation when fed orally by releasing a metabolite ricinoleic acid in the intestine and that interact with the prostanoid receptor EP_3 (Tunaru, 2012). This study was aiming to evaluate EEAA effect on reducing diarrheal activity. Treatment of EEAA (400mg/kg) showed a notable and dose-depended reduction (52.8%) of watery

stools. Loperamide (5 mg/kg) was used as standard in this experiment to validate the method and significantly reduced defecation rate (67%) (Figure 3).

Thrombolytic activity: Thrombolytic drugs dissolve blood clots by activating plasminogen, which is a proteolytic enzyme that is capable of break link between fibrin molecules. Here we studied EEAA for possible activity on plasminogen activation. Treatment with EEAA demonstrated remarkable (p<0.001) and dose-dependent dissolution of blood clot (45.2 ± 0.21%) compared to control (Figure 4). Streptokinase was the standard drug that showed 75 ± 0.09% clot lysis.

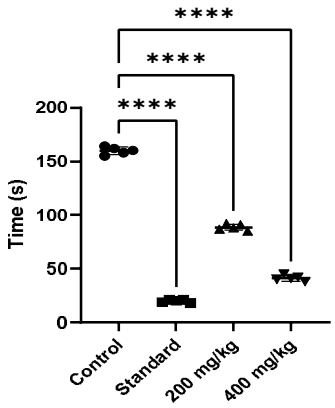

Figure 2. *In vivo* Tail suspension test (TST) of ethanol extract of *Amomum aromaticum* leaves.Values are expressed as mean ± SEM (n = 5); ****p< 0.0001 and are statistically significant compared to control followed by one-way ANOVA (GraphPad Prism 9). (EEAA) = Ethanol extract of *Amomum aromaticum.*

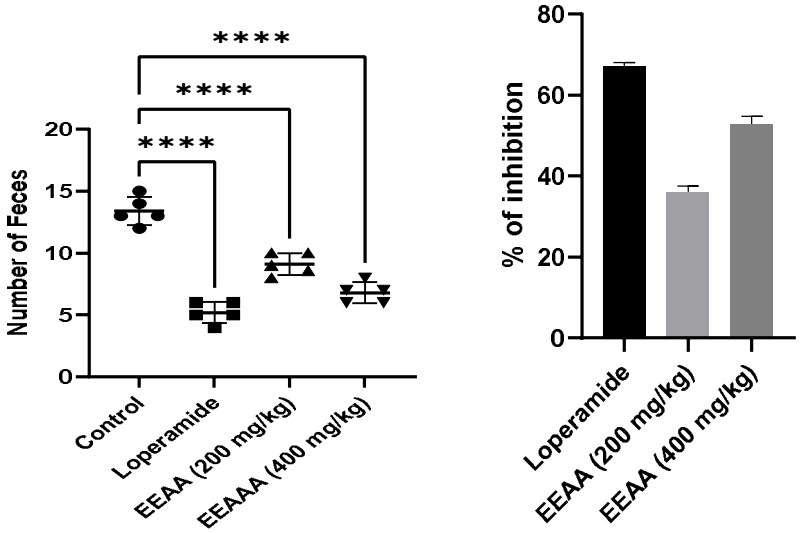

Figure 3. *In-vivo* anti-diarrheal effect of ethanol extracts of *Amomum aromaticum* on *Swiss albino* mice. Values are expressed as mean ± SEM (n = 5); ****p<0.0001 are statistically significant compared to control followed by Dunnet test (GraphPad Prism 8.4). (EEAA) =Ethanol extract of *Amomum aromaticum.*

Castor oil is a natural compound, containing triglyceride, ricinoleic acid, and induces laxation (IWAO and TERADA, 1962). When ingested, castor oil releases active metabolite ricinoleic acid, afterwards ricinoleic acid releases prostanoids from intestinal tissues and interact with EP_3 receptor of prostaglandin E_2 (PGE_2) (Capasso *et al.*, 1987). The anti-diarrheal activity shown by EEAA was the reversal of castor oil mechanism. The extracts meets some criteria that were similar with loperamide such as prevented the production of wet and formless feces also inhibited GIT propulsive act. It has been reported that as therapeutic effect of loperamide have anti-motility and anti-secretory activities (Baker, 2007; Hovdenak, 1987).

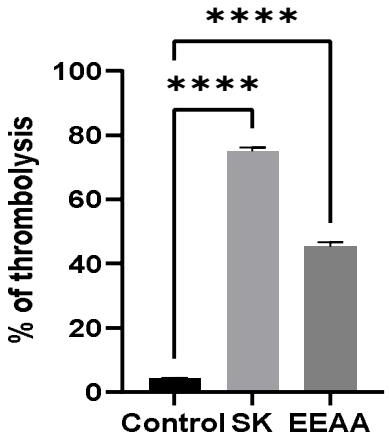

Figure 4. *In vitro* thrombolytic test of ethanol extract of *Amomum aromaticum* leaves. Values are expressed as mean ± SEM (n = 5); ****p<0.0001 are statistically significant compared to control followed by one-way ANOVA (GraphPad Prism 9). (EEAA) =Ethanol extract of *Amomum aromaticum.*

Although immobile period in FST represents major depression symptoms, but it can also be a form of apathy or avolition. Which could be partly related to negative symptoms like schizophrenia (Arauchi *et al*., 2018). As EEAA showed a potential anti-depressant activity in this current study, a comprehensive study could be direct in future to evaluate its effect in neuropsychiatric disorder also. The outcome of the current study proposes the anti-depressant activity of EEAA in FST as well as TST in animal models. The ethanol extract significantly diminished the immobility-period in both Force swimming and Tail suspension test. FST and TST both characterise the behavioral depression model, claimed to mimic a condition comparable to human despair (Borsini and Meli, 1988; Can *et al*., 2012). The FST were developing a depressive environment when positioned in an inescapable compartment. Instead, in the TST method the mice were hung along with tail, inducing depressive situation. The most important theory of mood-disorder is interrelated to the variations in the altitudes of biogenic-amines (Heninger *et al*., 1996; Maas, 1975; Miller *et al*., 1996). The event of depression is related with the changes in the levels of biogenic-amines into brain such as NE, dopamine (DA), 5-hydroxytryptamine (5-HT), indolamine, epinephrine and serotonin (Koslow *et al*., 1983). Pretreatment with EEAA (200 and 400 mg/kg) and fluoxetine showed significant *($p<0.001$)* inhibition of immobility period with dose dependency in FST and TST, compare with control described in figure 1 and 2. Fluoxetine is selective serotonin reuptake inhibitors (SSRIs) (Singh *et al*., 2001). It causes rapid increase of serotonin into somatodendritic region of serotonergic neurons which desensitises the somatodendritic serotonin-1A autoreceptors (BLIER *et al*., 1982). Desensitisation of postsynaptic serotonin receptors may contribute to the therapeutic actions of Fluoxetine (Foote and Morrison, 1987; Khushboo and Sharma, 2017). This bio effect of EEAA treated models may be due to its reducing effect in endogenous despair. Thus, the activity of EEAA may involve in the mechanisms of anti-depressant agents as described above.

Platelets play a vital role in developing atherothrombosis and damage regions of endothelial surface (Davì and Patrono, 2007). The stimulated platelets fabricate reticulation with each other than attach to leucocytes carrying them into a complex process of plaque formation. Plasmin is a fibrinolytic mediator, which cleave the fibrinogen and fibrin in a clot (Ablondi and De Renzo, 1959). Streptokinase like thrombolytic agent can repel the pathway of blood clot formation. The ultimate task of thrombolytic treatment is the degeneration of fibrin through plasmin, which can be initiated by dint of the activators from inactive plasminogen. Streptokinase activates plasminogen to dissolve clots (Reddy and Markus, 1972; A. J. Johnson and McCarty, 1959). By comparing with this positive (Streptokinase) and negative control, a significant *($p<0.001$)* thrombolytic bioactivity was detected after treating the blood clots with EEAA (Figure 4).

EEAA exhibited excellent thrombolytic activity by clot lysis relatively close to the standard candidate showed earlier. The chemical constituents existing in this respective extract may activate or stimulate the plasmin factor for initiating thrombolytic pathway. From this above outcome some observe that the plants extract revealed notable thrombolytic action.

Conclusions

The ethanol extract of *A. aromaticum* (EEAA) demonstrated a moderate anti-depressant, antidiarrheal and significant thrombolytic effects. Quantitative phytochemical characterisation and screening of the broad-spectrum pharmacological activity of EEAA need to be investigated.

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References

Ablondi, F. and De Renzo, E. 1959. Mechanism of clot lysis by streptokinase and effects of epsilon-aminocaproic acid. *Proc. Soc. Exp. Biol. Med.* **102**, 717-719.

Akinmoladun, A. C., Ibukun, E., Afor, E., Obuotor, E. M. and Farombi, E. 2007. Phytochemical constituent and antioxidant activity of extract from the leaves of Ocimum gratissimum. *Sci. Res. Essays.* ***2***, 163-166.

Arauchi, R., Hashioka, S., Tsuchie, K., Miyaoka, T., Tsumori, T. and Limoa, E. 2018. Gunn rats with glial activation in the hippocampus show prolonged immobility time in the forced swimming test and tail suspension test. *Brain Behav.* **8**, e01028.

Awoyinka, O., Balogun, I. and Ogunnowo, A. 2007. Phytochemical screening and in vitro bioactivity of Cnidoscolus aconitifolius (Euphorbiaceae). *J. Med. Plant Res.* **1**, 63-65.

Awuchi, C.G. 2019. Medicinal plants: the medical, food, and nutritional biochemistry and uses. *Int. J. Adv. Acad. Res.* **5**, 220-241.

Bahramsoltani, R., Farzaei, M. H., Farahani, M. S. and Rahimi, R. 2015. Phytochemical constituents as future antidepressants: a comprehensive review. *Rev Neurosci*. **26**, 699-719.

Baker, D. E. 2007. Loperamide: a pharmacological review. *Rev Gastroenterol Disord.* **7**, S11-18.

Blier, P., de Montigny, C. and Chaput, Y. 1987. Modifications of the serotonin system by antidepressant treatments: implications for the therapeutic response in major depression. *J. Clin. Psychopharmacol.* **7**, 24S.

Borsini, F. and Meli, A. 1988. Is the forced swimming test a suitable model for revealing antidepressant activity? *Psychopharmacol.* **94**, 147-160.

Can, A., Dao, D. T., Terrillion, C. E., Piantadosi, S. C., Bhat, S. and Gould, T. D. 2012. The tail suspension test. *JoVE J. Vis. Exp.* **59**, e3769.

Capasso, F., Tavares, I. A., Tsang, R., Rennie, J. A. and Bennett. 1987. Eicosanoid formation by mammalian intestine. Effects of some intestinal secretagogues. *Eur. J. Pharmacol.* **138**, 107-113.

Cryan, J. F. and Mombereau, C. J. M. p. 2004. In search of a depressed mouse: utility of models for studying depression-related behavior in genetically modified mice. *Mol. Psychiatry*. **9**, 326-357.

Dang, N. H., Anh, L. T. V. and Dat, N. T. 2020. Anti-Inflammatory Effects of Essential Oils of Amomum aromaticum Fruits in Lipopolysaccharide-Stimulated RAW264.7 Cells. *J. Food Qual.* **2020**, 8831187.

Davì, G. and Patrono, C. 2007. Platelet activation and atherothrombosis. *N. Engl. J. Med.* **357**, 2482-2494.

Dewan, S. M. R. and Das, A. 2013. Investigation of *in vitro* thrombolytic potential and phytochemical nature of Crinum latifolium L. leaves growing in coastal region of Bangladesh. *Int. J. Biol. Res.* **4**, 1-7.

Foote, S. L. and Morrison, J. H. 1987. Development of the noradrenergic, serotonergic, and dopaminergic innervation of neocortex. *Curr. Top. Dev. Biol.* **21**, 391-423.

Gakunga, N. J., Kateregga, G., Sembajwe, L. F. and Kateregga, J. 2013. Antidiarrheal activity and phytochemical profile of the ethanolic leaf extract of *Leonotis nepetifolia* (Lion's ear) in Wistar albino rats. *J. Complement. Med. Research.* **2,** 121-126.

Ganapaty, S., Dash, G., Subburaju, T. and Suresh, P. 2002. Diuretic, laxative and toxicity studies of *Cocculus hirsutus* aerial parts. *Fitoterapia.* **73,** 28-31.

Ghosh, A., Banik, S., & Islam, M. A. J. B. J. o. P. 2015. In vitro thrombolytic, anthelmintic, anti-oxidant and cytotoxic activity with phytochemical screening of methanolic extract of *Xanthium indicum* leaves. *Bangladesh J. Pharmacol.* **10,** 854-859.

Heninger, G., Delgado, P. and Charney, D. 1996. The revised monoamine theory of depression: a modulatory role for monoamines, based on new findings from monoamine depletion experiments in humans. *Pharmacopsychiatry.* **29,** 2-11.

Hossain, S., Sarkar, B., Prottoy, M. N. I., Araf, Y., Taniya, M. A. and Ullah, M. A. 2019. Thrombolytic activity, drug likeness property and ADME/T analysis of isolated phytochemicals from ginger (zingiber officinale) using *in silico* approaches. *Mod. Res. Inflamm.* **8**, 29-43.

Hovdenak, N. 1987. Loperamide treatment of the irritable bowel syndrome. *Scand. J. Gastroenterol.* **22,**81-84.

Islam, A., Hussain, M. S., Sen, N., Abedin, F., Millat, M. S., Islam, Mohammad S. and Hossain, M. M. 2017. Investigation of in vitro thrombolytic and anti-helminthic activity and *in vivo* anxiolytic and antidepressant potentiality with phytochemical nature of methanolic extract of *Leucas lavandulifolia*. *Sustain. Chem. Pharm.* **6**, 61-66.

Iwao, I. and Terada, Y. 1962. On the mechanism of diarrhea due to castor oil. *Jpn. J. Pharmacol.* **12,** 137-145.

Johnson, A. J. and McCarty, W. R. 1959. The lysis of artificially induced intravascular clots in man by intravenous infusions of streptokinase. *J. Clin. Invest.* **38,** 1627-1643.

Johnson, P. C., Ericsson, C. D., DuPont, H. L., Morgan, D. R., Bitsura, J. A. M. and Wood, L. V. J. J. 1986. Comparison of loperamide with bismuth subsalicylate for the treatment of acute travelers' diarrhea. *JAMA.* **255,** 757-760.

Kala, C. P. 2005. Ethnomedicinal botany of the Apatani in the Eastern Himalayan region of India. *J. Ethnobiol. ethnomed.* **1**, 11.

Khushboo, S. B. and Sharma, B. 2017. Antidepressants: mechanism of action, toxicity and possible amelioration. *J. Appl. Biotechnol. Bioeng,* **3,** 1-13.

Koslow, S. H., Maas, J. W., Bowden, C. L., Davis, J. M., Hanin, I. and Javaid, J. 1983. CSF and urinary biogenic amines and metabolites in depression and mania: A controlled, univariate analysis. *Arch. Gen. Psychiatry.* **40,** 999-1010.

Maas, J. W. 1975. Biogenic amines and depression: Biochemical and pharmacological separation of two types of depression. *Arch. Gen. Psychiatry.* **32**, 1357-1361.

Martins, J. and S, B. 2018. Phytochemistry and pharmacology of anti-depressant medicinal plants: A review. *Biomed. pharmacother.***104**, 343-365.

Miller, H. L., Delgado, P. L., Salomon, R. M., Berman, R., Krystal, J. H., Heninger, G. R. and Charney, D. S. 1996. Clinical and biochemical effects of catecholamine depletion on antidepressant-induced remission of depression. *Arch. Gen. Psychiatry.* **53**, 117-128.

Rahman, M. K., Chowdhury, M. A. U., Islam, M. T., Chowdhury, M. A., Uddin, M. E. and Sumi, C. D. 2015. Evaluation of Antidiarrheal Activity of Methanolic Extract of Maranta arundinacea Linn. Leaves. *Adv Pharmacol Sci*. **2015**, 257057.

Reddy, K. N. N. and Markus, G. 1972. Mechanism of activation of human plasminogen by streptokinase Presence of active center in streptokinase-plasminogen complex. *J. Biol. Chem.* **247,** 1683-1691.

Richelson, E. 1982. Pharmacology of antidepressants in use in the United States. *J. Clin. Psychiatry.* **43**, 4.

Sharma, P. K., Chauhan, N. S. and Lal, B. 2004. Observations on the traditional phytotherapy among the inhabitants of Parvati valley in western Himalaya, India. *J. Ethnopharmacol*. **92,** 167-176.

Shoba, F. G. and Thomas, M. 2001. Study of antidiarrhoeal activity of four medicinal plants in castor-oil induced diarrhoea. *J. Ethnopharmacol.* **76**, 73-76.

Singh, V. P., Jain, N. K. and Kulkarni, S. 2001. On the antinociceptive effect of fluoxetine, a selective serotonin reuptake inhibitor. *Brain Res.* **915**, 218-226.

Steru, L., Chermat, R., Thierry, B. and Simon, P. 1985. The tail suspension test: a new method for screening antidepressants in mice. *Psychopharmacol.* **85,** 367-370.

Toyin, Y. M., Khadijat, O. F., Saoban, S. S., Olakunle, A. T., Abraham, B. F. and Luqman, Q. A. 2012. Antidiarrheal activity of aqueous leaf extract of Ceratotheca sesamoides in rats. *Bangladesh J. Pharmacol.* **7**, 14-20.

Tunaru, S., Till F.A., Rolf M. Nüsing, b Martin D. and Stefan Offermanns. 2012. Castor oil induces laxation and uterus contraction via ricinoleic acid activating prostaglandin EP3 receptors. *Biol. Sci*. **109,** 9179-9184.

Zomkowski, A. D., Santos, A. R.. and Rodrigues, A. L. 2005. Evidence for the involvement of the opioid system in the agmatine antidepressant-like effect in the forced swimming test. *Neurosci. Lett.* **381,** 279-283.

Zomkowski, A. D. E., Rosa, A. O., Lin, J., Santos, A. R., Calixto, J. B. and Rodrigues, A. L. S. 2004. Evidence for serotonin receptor subtypes involvement in agmatine antidepressant like-effect in the mouse forced swimming test. *Brain Res.* **1023**, 253-263.